The specification has been amended to make reference to the PCT from which this case derives. Reference is also made to the priority application.

Pursuant to the Examiner's request, submitted herewith is a substitute specification. The specification provided herewith is merely a clean copy of the application as would have been transmitted by the International Bureau.

The claims have been revised to define the invention with additional clarity. The claims as presented are fully supported by an enabling disclosure. The revision of claim 1 finds support, for example, in now cancelled claim 5 and the revision of claim 3 finds support, for example, at page 11, lines 15-19. That the claims have been revised should not be construed as an indication that Applicants agree with any view expressed by the Examiner. Rather, the revisions are made merely to advance prosecution and Applicants reserve the right to pursue any deleted subject matter in a continuation application.

Claims 10-12, 16-17 and 20 stand rejected under 35 USC § 101 as encompassing natural phenomena. Withdrawal of the rejection is submitted to be in order in view of the above-noted claim revisions and comments that follow.

Claims 10, 11, 13, 14 and 18 have been amended to refer to plant cells that contain a heterologous nucleic acid construct. Such cells are not found in nature.

Claim 20 has been cancelled thereby rendering moot the rejection thereof.

Reconsideration is requested.

Claims 6 and 20 stand rejected under 35 USC §112, second paragraph, as allegedly being indefinite. Claim 6 has been amended as suggested by the Examiner and claim 20 has been cancelled. Reconsideration is requested.

Claims 1 and 3-20 stand rejected under 35 USC § 112, first paragraph, as allegedly being non-enabled. Withdrawal of the rejection is believed to be in order for the reasons that follow.

The Examiner contends that the promoter function of sequences or sequence fragments of Badnavirus strains is unpredictable because strain differentiation is unpredictable and the skilled person would therefore be required to engage in undue experimentation to practice the invention as claimed. Applicants respectfully disagree.

Cabauatan et al teaches the phylogenetic relationship between variants of rice tungro bacilliform virus (RTBV), which is distinct from other Badnaviruses in having four ORFs. The Examiner indicates that strain variants may have

low sequence homology because of the 'quasi-species genetic population structure' of RTBV.

The six RTBV variants are 'between 95 and 99% identical at the nucleotide and amino acid levels' (Abstract line 4 and Table 2). Thus, they show high levels of sequence identity, despite containing substitutions, deletions and insertions compared to the published RTBV sequence.

Cabauatan et al at page 2236, column 2, lines 10-22, teaches that studying genomic sequences within a heterogeneous viral population (i.e. a quasi-species) is useful for studying adaptive or evolutionary changes within the virus. It does not follow that a 'large sampling of quasispecies genomic structure would be required to determine sequences and their functional uses' as alleged by the Examiner. The genomic sequences not only possess high overall levels of identity as described above but also possess 'highly conserved regions' (page 2236, last line). Given these high levels of identity, an artisan could readily identify key functional regions in different strains or variants of BSV. Large scale sampling would be unnecessary.

The existence of a 'quasi-species genetic population structure' does not mean that promoter function is

inherently unpredictable as alleged by the Examiner. As described in Regenmortel et al, the term 'quasispecies' has a meaning broadly in line with what a biologist would term a species, i.e., a population of related individuals each having a variant sequence. Indeed, the term 'species' is commonly used to describe these families (see enclosed Plant Virus Online description of Badnavirus). The term 'quasispecies' is used to distinguish such a population from a chemical 'species', all the members of which are identical. An RNA quasispecies, for example, is a heterogeneous population of RNA molecules of varying sequence rather than a population of RNA molecules of identical sequence (see page 9, middle paragraph). Thus, there is nothing unusual about a 'quasispecies genetic population structure' compared to the genetic population structure of any other organism, which makes the identification of corresponding functional elements more problematic in different members of the population.

The Examiner further asserts that chemical hybridization is unpredictable in identifying a Badnavirus species, on the basis of the teaching of Lockhart et (1993).

Lockhart et al (1993) discusses various approaches for the detection of BSV. One of these approaches is dot-blot

hybridization, which involves the immobilization of genomic DNA from one BSV isolate on a membrane and hybridization with labelled genomic DNA from a different isolate (which may be PCR amplified). Under the hybridization conditions employed, no cross-hybridization was reported between BSV isolates, and only one BSV isolate hybridized to any of the SCBV isolates (this data is not shown). Dot-blot analysis using known BSV genomic DNA under the conditions used in Lockhart et al is thus found to be unsuitable for identifying an isolate as BSV.

Various techniques for the typing of viruses, including visual inspection, electron microscopy, dsRNA detection, serotyping and dot blotting are found by Lockhart et a to be unsuitable for BSV detection. However, a PCR based technique (which involves the chemical hybridization of primer sequences to conserved target sequences) is used successfully to detect BSV isolates in Lockhart et al.

In the present claims, hybridization conditions are recited to define a genus of nucleic acid molecules that consist of a promoter sequence active in Musaceae spp. The hybridization of two nucleic acid molecules under a particular set of conditions is dependent on the extent of complementary between those particular molecules. This is

an entirely predictable process. Under low stringency conditions, hybridization will occur between molecules with low complementarity and under high stringency conditions, hybridization will occur only between molecules that have high complementarity.

While Lockhart et al teaches that under certain hybridization conditions the genomes of BSV isolates do not hybridize (i.e., do not possess the required level of sequence complementarity or identity), there is no teaching regarding the hybridization or otherwise of isolated functional elements within the BSV genome. The hybridization of such elements under particular conditions is predictable and is entirely dependent on the degree of sequence identity between the isolated elements. Irrespective of the suitability of dot blot analysis for detecting BSV strains, chemical hybridization is an effective and entirely predictable method for establishing that one nucleic acid molecule (e.g., an isolated promoter element) is closely related to another nucleic acid molecule (i.e., shares high sequence identity with it).

The Examiner alleges that the functional promoter capabilities of a polynucleotide as claimed are unpredictable for conferring expression in all plant cells in all tissue types on the basis of Olszewski et al, and in

particular on the alleged absence of ScBV promoter driven GUS expression in non-vascular leaf cells and awn tissue. The teaching of Olszewski et al is entirely directed at the SCBV promoter region and does not concern the BSV promoter.

Expression in the Olszewski et al experiments is detected by histochemical staining of plant tissue. Expression below the detection threshold does not lead to staining and so produces a negative result in this assay. However, even low level expression may be sufficient for many purposes.

Although the Examiner asserts that there is no expression in non-vascular tissue, page 38, line 28-29, clearly indicates that some expression is in fact observed in the non-vascular leaf tissue. Furthermore, references to vascular expression in the tables (for example sepal and silique tissue in Table 6) is not a teaching that there was no expression in non-vascular tissue, merely that the levels of expression recorded in the Tables only relate to expression levels in the vascular tissue.

In Olszewski et al, ScBV promoter driven expression was detected in all tissues tested in stably transformed independent lines of *A sativa* (Table 2), in all the tissue tested of *A. sativa* T1 seedlings (Table 3), in 10 of the 11 tissues tested in *A sativa* T1 plants (Table 4), in

expression in 6 of 6 tissues tested in *A. thaliana* T1 plants (Table 5) and in expression in 8 of 8 tissues tested in *A. thaliana* T2 plants (Table 6).

Actually, the teaching of Olszewski et al to the skilled artisan would be that the ScBV promoter is a constitutive promoter that drives gene expression almost ubiquitously (see table 2) in the tissues of a range of different plant spp.

The absence, in T1 oats, of expression in the awn, which is a hair-like bristle extending from the upper part of the lemma of certain plants, does nothing to alter this teaching.

Furthermore, there is no reason to expect, in the light of the teaching of Olszewski et al, that BSV promoter-driven expression would not occur in a wide range of plant species. Claims covering plant cells and plants are therefore fully enabled.

Chen et al teaches the presence of an enhancer element downstream of the transcriptional start site of RTBV. RTBV is the most divergent member of the Badnavirus family. It has about 40% sequence similarity with BSV, four ORFs and is transmitted only with rice turgo spherical virus (RTSV) by leafhoppers, unlike other Badnaviruses which are transmitted by mealybugs. In contrast to the CaMV

promoter, Chen et al indicates that the RTBV promoter sequence requires an enhancer element in order to direct high levels of expression.

However, the data in the present specification (Example 2, Table 4) clearly show that the BSV promoter described is sufficient to drive high levels of reporter gene expression. While the activity of the promoter may, as with any promoter, be affected by the presence of other regulatory elements, the promoter sequence as defined is itself sufficient to drive expression. It is irrelevant to speculate on whether the addition of other regulatory elements might alter expression in some way - Applicants have shown that the promoter itself has the claimed activity and this is sufficient to meet the enablement requirement of USC 35 §112, first paragraph.

It is further noted that the promoter region described in the present specification (SEQ ID NO:2) corresponds to -350 to +100 relative to the transcription start site (page 4 lines 24-25). If an enhancer corresponding to that of RTBV were present in BSV, it would be included within the present construct. There is no evidence to suggest that other regulatory elements outside this region can affect BSV promoter-driven expression.

Hao et al and Izawa et al teach that the modification of key residues in regulatory motifs can affect the binding of transcription factors. Based on this teaching, the Examiner alleges that no guidance is provided for which bases of SEQ ID NO:2 can be altered to maintain activity.

The present claims are directed at polynucleotides that are promoter sequences of isolates of BSV. Thus, while potentially being allelic variants of SEQ ID NO:2, the claimed promoter sequences are necessarily active, as a virus with an inactive promoter could not infect a source and replicate to produce a population from which isolates could be made. The present specification thus teaches the skilled person how to identify active sequence variants and an artisan could obtain and use an active promoter without any knowledge of which residues contribute to this activity. There is no requirement to modify individual residues and test for activity, although such routine experimentation is entirely within the capability of the skilled person.

Furthermore, the sequence of the promoter as claimed is structurally defined in terms of its source, relationship to SEQ ID NO:2 and function. A promoter sequence must be functional and must have the sequence of SEQ ID NO:2, a sequence at least 75% similar to SEQ ID NO:2

or a sequence which hybridizes to SEQ ID NO:2 under stringent conditions. These are sufficient to define the active sequence and it is unnecessary to recite additional features, such as a specific length.

The totality of starting materials for producing transgenic plants and plant cells are fully discussed in the specification (for example, on page 15, line 12, to page 16, line 16,and page 17, line 27, to page 21, line 9). Of course, such materials and methods are well-known in the art and an artisan could readily make and use the invention as covered by these claims based on the teachings of the specification.

The present specification provides guidance for expression within Musaceae cells and Musaceae plants. As described above, Olszewski et al teaches the skilled person that the related SCBV promoter directs expression almost ubiquitously in plant tissue further, that the promoter is active in a wide variety of plant species (including *A. sativa, A thaliana* and *N. tabacum*). The apparent absence of detectable expression from the awn of T1 *A. sativa* plants would not teach a skilled person that the ScBV promoter is limited to specific tissues, viewed in light of the other data disclosed in Olszewski et al.

Given the teachings of the specification, no undue experimentation would be required to make and use the invention as claimed. Contrary to the Examiner's assertion, active BSV promoter sequences that are allelic variants of SEQ ID NO:2 could be obtained by a skilled person and used as described without any undue burden of experimentation.

Reconsideration is requested.

Claims 1 and 3-20 stand rejected under 35 USC § 112, first paragraph, as allegedly lacking written description. The rejection is traversed.

The invention as claimed is described by clear and unique structural features, including the source of the element.

The specification indicates that the key feature of the claimed invention is the isolated nucleic acid that is a promoter element from a BSV isolate that consists of SEQ ID NO:2, hybridizes to SEQ ID NO:2 under highly stringent conditions or is at least 75% similar to SEQ ID NO:2, or a fragment thereof that directs expression in one or more cells of a Musaceae plant.

The source of the promoter element is a clear structural feature. Despite the existence of heterogeneity within each species, BSV and ScBV are distinct species of Badnavirus. This is evident from the art, which

consistently lists them as separate species (see for example, 'Badnavirus' from Plant Viruses Online; enclosed). Indeed, at the amino level, BSV is more closely related to CoYMV than ScBV (Harper and Hull (1998) of record: page 277 1st col). The designation of a nucleic acid sequence as a BSV sequence is thus a structural feature which distinguishes the sequence in the same way as the designation of a nucleic acid sequence as a mammalian or human sequence.

Claims are drawn to a genus of polynucleotides that are promoter elements from BSV isolates that consist of SEQ ID NO:2, hybridize to SEQ ID NO:2 under highly stringent conditions or are at least 75% similar to SEQ ID NO:2, or fragments of these which direct expression in one or more cells of a Musaceae plant.

The present promoter sequence is thus defined in structural as well as functional terms. The claimed genus is thus defined in terms of what the promoter is, as well as what it does, and the written description requirements set out in University of California v Eli Lilly and Co are thus fulfilled.

The specification discloses a single species (a molecule consisting of SEQ ID NO:2) that is within the

scope of the claimed genus. There is actual reduction to practice of the disclosed species.

Hybridization techniques using a known DNA as a probe under given conditions were conventional in the art at the time of filing, as taught, for example on page 10, line 5, to page 12, line 4, of the specification. Also well known were methods of determining similarity/homology, as taught on page 9, lines 3-28, of the specification, as methods of determining promoter activity, as taught on page 6, line 27, to page 7, line 11, and Example 2.

A person of skill in the art would thus be provided with all the means to identify and obtain other species within the genus. While the sequences of BSV may contain natural variations, the stringent criteria of the claims in terms of source, hybridization conditions and percentage homology would yield structurally similar DNAs, and there would be no substantial variation among species that are within the scope of the claims.

Thus, a representative number of species is disclosed, since highly stringent hybridization conditions in combination with the activity of DNA and the level of skill and knowledge in the art are adequate to determine that Applicants were in possession of the claimed invention. Reconsideration is thus requested.

Claims 3-4 and 7 stand rejected under 35 USC 102(b) as allegedly being anticipated by Lockhart et al and Lockhart et al. The rejections are traversed.

Both the above Lockhart references describe dot blot hybridization with genomic extracts from BSV isolates and PCR amplified products thereof.

The isolates of Lockhart et al contain the whole BSV genome, including ORFs. There is no disclosure in either Lockhart reference of 'an isolated polynucleotide consisting of a promoter sequence' as required by claim 3. This wording excludes the whole BSV genome and other polynucleotides that comprise a promoter along with additional sequence, for example coding sequence.

Claim 4 is dependent on claim 3 and claim 7, by virtue of its dependency on claim 6 is required to contain non-BSV sequence. The whole BSV genome is excluded.

Claims 3-4 and 7 are thus novel over Lockhart et al (1986) and Lockhart et al (1993) and reconsideration is requested.

Claims 1 and 3-20 stand rejected under 35 USC 102(e) as allegedly being anticipated by Olszewski et al in light of Lockhart and Regenmortel. The rejection is traversed.

Olszewski et al discloses the use of promoter sequences from SCBV. The Examiner contends that a range of

factors 'confound the designation of the pararetroviruses known as BSV or ScBV' and the two 'species' represent 'subsets of quasispecies of a population of Badnavirus with overall indistinguishable characteristics'. Thus, the ScFV sequences of Olszewski are considered to anticipate the BSV sequences disclosed.

The criteria cited in Regenmortel for species demarcation are:

1) Differences in host ranges

2) Differences in genome nucleotide sequences of more than 50%

3) Differences in gene product sequences

Lockhart et al indicates that the host range of BSV is restricted to banana and sugarcane, overlapping to some extent with ScBV.

Table 1 of the specification shows that ScBV has 51.2 % sequence 'homology' with BSV. 'Homology' is defined on page 9, line 16, of the specification as similarity. These are terms commonly used in the art. Sequence similarity/homology differs from sequence identity because it takes conservative substitutions into account. Thus, two sequences can be similar at a position if the two residues at that position have similar chemical properties, even if

they are actually different residues (i.e., there is similarity but not identity).

The Regenmortel et al criteria refer to differences in sequence of greater than 50%. This wording indicates that conservative substitutions are considered to be differences. While ScFV may have 51.2 % similarity to BSV, the % identity is less than 50% and therefore the two species meet this criteria for demarcation. In terms of gene products, BSV is more closely related to CoYMV (Commelina yellow mottle virus) than ScBV (see Harper and Hull (1998) of record: page 274 line 12 and Table 1).

Thus, in at least two of the three Regenmortel criteria, ScBV and BSV are clearly shown to be different species. There is no basis in the art for the Examiner's assertion that ScBV and BSV are the same species.

The distinction between ScBV and BSV finds support in the prior art, which consistently refers to ScBV and BSV as being different species. References to members of the Badnavirus class, for example list both species separately (see for example Lockhart et al (1993) page 106, 'Badnavirus': Plant Virus Online, filed herewith). Furthermore, while difficulties have been encountered in detecting heterogeneous populations of BSV, specific detection of BSV has been shown using PCR (Lockhart et al

(1993) and immuno-capture-PCR (IC-PCR: present specification). An isolate of BSV is therefore distinct from an isolate of ScBV.

The ScBV sequences disclosed in Olewski et al do not hybridize to SEQ ID NO:2 under the conditions recited in the claim 3. Furthermore, none of these sequences shows greater than 75% similarity with SEQ ID NO:2 as required by claim 1. The ScBV sequences in Olewski et al thus fall outside the present claims and therefore the reference is not anticipatory. Reconsideration is requested.

Attached hereto is a marked-up version of the changes made to the claims by the current amendment. The attached pages are captioned "**Version With Markings To Show Changes Made**."

This application is submitted to be in condition for allowance and a Notice to that effect is requested.

Respectfully submitted,

NIXON & VANDERHYE, P.C.

By Mary J. Wilson
Mary J. Wilson
Reg. No. 32,955

MJW:tat

1100 North Glebe Road, 8th Floor
Arlington, Virginia 22201-4714
Telephone: (703) 816-4000
Facsimile: (703) 816-4100

VERSION WITH MARKINGS TO SHOW CHANGES MADE

IN THE CLAIMS:

1. (Amended) An isolated polynucleotide consisting of a Banana Streak Virus promoter sequence selected from:

(a) the promoter sequence of a Nigerian isolate of Banana Streak Virus shown in SEQ ID NO. 2;

(b) a promoter sequence of an isolate of Banana Streak Virus, which promoter sequence is an allelic variant of the promoter sequence of SEQ ID NO. 2; wherein said allelic variant has a sequence that is at least 75% identical to the promoter sequence of SEQ ID NO:2,

(c) a fragment of (a) or (b) which, when operably linked to a transcribable sequence, promotes transcription of the transcribable sequence in one or more cells of a *Musaceae* plant [cell].

2. (Amended) An isolated polynucleotide [according to claim 1 wherein said] consisting of a Banana Streak Virus promoter having the sequence [is] shown in SEQ ID NO. 2.

3. (Amended) An isolated polynucleotide consisting of a promoter sequence, which promoter sequence:

(i) selectively hybridizes under stringent conditions with a polynucleotide complementary to a polynucleotide which has the nucleotide sequence shown in SEQ ID NO: 2;

wherein said stringent conditions comprise overnight hybridization at 42°C in 0.25M Na_2HPO_4, pH 7.2, 6.5% SDS, 10% detran sulfate and a final wash at 55°C in 0.1x SSC, 0.1% SDS, and;

(ii) when operably linked to a transcribable sequence promotes transcription of the transcribable sequence in one or more cells of a *Musaceae* plant [cell].

4. (Amended) An isolated polynucleotide according to claim [2] 3 which has a nucleotide sequence found in a strain of Banana Streak Virus.

6. (Twice Amended) A nucleic acid construct comprising the promoter sequence of a polynucleotide according to any one of claims 1, 3 and 5, and a non-Banana Streak Virus transcribable sequence.

7. (Amended) A nucleic acid construct according to claim 6 wherein the promoter sequence is operably linked to [a] the transcribable sequence.

8. (Twice Amended) A nucleic acid vector suitable for transformation of a plant cell and including a nucleic acid construct according to claim 6 [the promoter sequence of a polynucleotide according to any one of claims 1, 3 and 5].

10. (Twice Amended) A plant cell containing a heterologous [polynucleotide,] nucleic acid construct [or nucleic acid vector] according to [any one of claims 1, 3 and 5] claim 6.

11. (Amended) A plant cell according to claim 10 wherein said plant cell has said [having a] heterologous [said polynucleotide or] nucleic acid construct within its chromosome.

13. (Twice Amended) A method of producing a cell according to claim 10, the method including incorporating said [polynucleotide,] nucleic acid construct [or nucleic acid vector] into the cell by means of transformation.

14. (Amended) A method according to claim 13 which includes recombining [said polynucleotide or]said nucleic acid construct with the cell genome nucleic acid such that it is stably incorporated therein.

18. (Twice Amended) A method of producing a plant, the method including incorporating <u>the</u> [a polynucleotide,] nucleic acid construct [or nucleic acid vector] according to [any of claims 1, 3 and 5] <u>claim 6</u> into a plant cell and regenerating a plant from said plant cell.